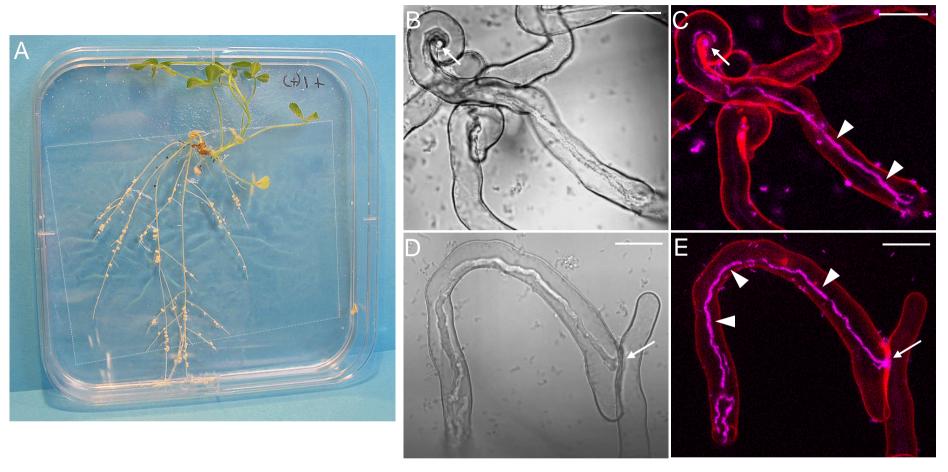
## Supplemental Figure S1



## Supplemental Figure S1 : Experimental set-up for *in vivo* confocal microscopic observation of rhizobial infection in *M. truncatula* root hairs.

**A**, For inoculation and subsequent microscopic observation, *M. truncatula* plants were placed on semi-solid medium in Petri dishes, and the root system covered with BioFolie film (indicated with the dashed line). A supernodulated *M. truncatula sunn* composite plant is shown 2 weeks after inoculation with *S. meliloti* 2011. Note the large number of nodules which have developed under the film.

**B-E**, Mature infection sites in *M. truncatula* root hairs growing under BioFolie film. Bright field images are shown on the left (**B**, **D**), and the corresponding confocal images showing both the cCFP fluorescence of the rhizobia (magenta) and cell wall autofluorescence (red), are on the right (**C**, **E**). Infection threads can initiate (arrows) either within curled root hairs (**B**, **C**), or between 2 appressed root hairs (**D**, **E**). Bacteria are often present in multiple files (arrowheads) within segments of mature threads (**C**, **E**). A significant proportion of the root hairs grow directly underneath and in close contact with the BioFolie film, thus greatly facilitating their observation in the xy plane. Bars =  $20 \,\mu m$